# What limits meiotic crossovers?

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Meiotic crossovers are points of exchange between homologous chromosomes that create genetic diversity in gametes. However, the number of crossovers (COs) is tightly constrained. At the lower limit, there is at least one CO per pair of homologous chromosomes per meiosis, and this "obligate" CO is essential for the balanced distribution of chromosomes in the gametes. At the upper end, the number of COs is relatively low in most eukaryotes, typically in the range of one to four per pair of chromosomes. This is rather surprising knowing that DNA double-strand breaks, which are the precursors of COs, occur in large excess (e.g., 20-fold in Arabidopsis) at the beginning of meiosis. What prevents most of the DSBs from becoming COs? We tackled this question by looking for factors which limit CO formation in *Arabidopsis thaliana*.[1]

The identification of genes that control CO formation has been very successful in a range of species over the past 25 y,[2] leading to a tremendous increase in our understanding of this process. However, almost all of the characterized genes promote CO formation. This is largely due to the fact that loss of pro-crossover activity, which leads to the loss of the obligate crossover, provokes a strong reduction in sporulation/fertility, which is readily detectable, thus facilitating the identification of the causal genes. On the contrary, very few anti-crossover activities are known, likely due to the lack of easy phenotypes to screen. To overcome this limitation we performed a screen in a context where an increase in crossovers was expected to be easily detectable. The trick was to start with a crossover-defective mutant with consequently reduced fertility and to look for mutations that restore fertility as a proxy for increased crossovers.

An advantage of Arabidopsis is that fertility is very easy to score (fruit size). Using this approach we identified the Arabidopsis homolog of Fanconi anemia complementation group M (FANCM) as a major meiotic anti-crossover factor. The FANCM helicase was previously identified due to its role in genome stability and DNA repair in yeasts and humans.[3] In humans, Fanconi anemia patients suffer a high predisposition to cancer and early-onset bone marrow failure. While no meiotic function of FANCM was previously described, meiotic recombination in Arabidopsis *fancm* mutants were increased by an unprecedented factor of almost 3-fold.[1] A parallel study also established a function of Arabidopsis FANCM in regulating meiotic crossover formation but described an allele that seemed to have a lesser effect.[4] Further independent work in fission yeast also demonstrated a meiotic anti-crossover activity of the FANCM homolog (Fml1), thus demonstrating the conservation of this function throughout kingdoms.[5] Hence, it will be interesting to decipher what is shared, beyond FANCM, between the Fanconi pathway of somatic DNA repair (which involves at least 14 other proteins in human) and the control of meiotic CO formation. In addition to the fundamental interest, the increase in CO frequency that we described in *Atfancm* has obvious implications for breeding programs, where crossovers are often limiting.

In many species, two pathways of CO formation co-exist, which have differing genetic requirements.[2] The first pathway (class I) produces interfering COs, a phenomenon where one CO prevents the formation of another nearby by an elusive mechanism.[6] The second pathway (class II) produces independently distributed crossovers. Interestingly, mutation of FANCM unleashed formation of class II COs (which is normally minor), while the first class appears to be untouched.[1] Therefore, it is still unknown what limits the number and controls the distribution of class I COs, which is the major class in budding yeast, mammals and plants. Further, even though the increase of COs in *fancm* is remarkable (~10 in wild-type vs. ~30 in *fancm*), the number is still far from the potential given by the number of double-strand breaks (~200). This strongly suggests that other strong anti-crossover activities must co-exist with FANCM. Two other meiotic anti-crossover activities have been described in eukaryotes and, intriguingly, like FANCM, they are both (different) helicases (*Sgs1*[7,8] in budding yeast and *RTEL-1*[9] in *C. elegans*) and have homologs in Arabidopsis. Combining mutations in different anti-CO activities, notably these helicases, may provide insight into the constraints on CO formation. Indeed, many species have only one to four crossovers per chromosome, which could have suggested a physical limit. However, the biggest chromosome in budding yeast, which measures 1.5 Mb, has an uncommon average of 10 CO per meiosis,[10] and is transmitted perfectly through generations. With such a CO density per unit of physical size, Arabidopsis chromosome 1 would receive 200 COs per meiosis, and the human chromosome 1 would receive 1,660 CO per meiosis. In addition, our result shows that the number of crossovers can be increased in Arabidopsis (from ~2 to ~6 per chromosome) with no detectable mechanical defects in chromosome segregation at meiosis. This suggests that there is an evolutionary force, which is not only mechanical, constraining the number of possible COs.

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